

platelet number was 198000 in high, and 213000 in low-intermediate group. Out of 35 patients who were checked for both HCV RNA and Anti-HCV 68.5% (n=24) positive for both Anti-HCV and HCV RNA, while 31.5% were positive for anti-HCV but HCV RNA negative. These patients are believed to be immune to HCV.

Conclusion: With the introduction of HCV quantification method as a diagnostic tool, it became possible to distinguish truly HCV immune patients who were considered as HCV infected before. We noticed positive correlation HCV viral load with liver enzymes. The method allows treatment monitoring for the first time in Mongolia.

PP-144 Influence of predictor variables on side effects of the treatment with PEG Interferon Alfa 2a plus ribavirin in chronic hepatitis C

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Aim: Prediction of main side effects of chronic hepatitis C treatment with Peg-IFN α2a and ribavirin.

Methods: We evaluated in 55 patients treated with Peg-IFN α-2a plus ribavirin numerous predictors influence on main side effects (dichotomous dependent variable) on different date of therapy: flu-like syndrome, leucopenia, thrombocytopenia, weight loss, fatigue, depression, insomnia, arthralgia, myalgia, anorexia, nausea, anemia, headache, alopecia, pruritus and rash. Statistic analyses was done by SPSS 11.0 with binary logistic regression. Eleven independent variables, such as age, gender, body mass index (BMI), disease limitation, pretreatment with standard or Peg-IFN, presence of antibodies to HBV, alcohol abuse, drug abuse, genotype (1b, 1a, 2 and 3), level of ferritin (FERR), viral load (VL), were coding as dichotomous or categorical.

Results: Already on first month of the treatment probability of myalgia calculated by formula 1:

$$\log \frac{p}{1-p} = 1.33 - 1.34 \text{HBV}(1) - 3.32 \text{disease limitation}(1) - 2.64 \text{disease limitation}(2), \quad (1)$$

where HBV(1) – HBcAb; disease limitation(1) – long-standing of HCV-infection up to 5 years; disease limitation(2) – 5–10 years. Probability of flu-like syndrome on first month of therapy is calculated by formula 2:

$$\log \frac{p}{1-p} = -2.54 + 2.69 \text{genotype 1a} + 8.07 \text{genotype 2} + 2.38 \text{genotype 3} + 2.67 \text{high VL} - 1.94 \text{alcohol abuse}. \quad (2)$$

Risk factors were genotype 1a (OR=14.7) and high VL (OR=1.45).

Significant model was received for IVDU (p=0.005), alcohol abuse, HBV infection, high FERR and VL influence on weight loss during last months of therapy (formula 3):

$$\log \frac{p}{1-p} = 1.73 - 4.36 \text{IVDU} - 1.78 \text{high FERR} - 0.038 \text{high VL} - 0.096 \text{alcohol abuse} + 1.55 \text{HBcAb}. \quad (3)$$

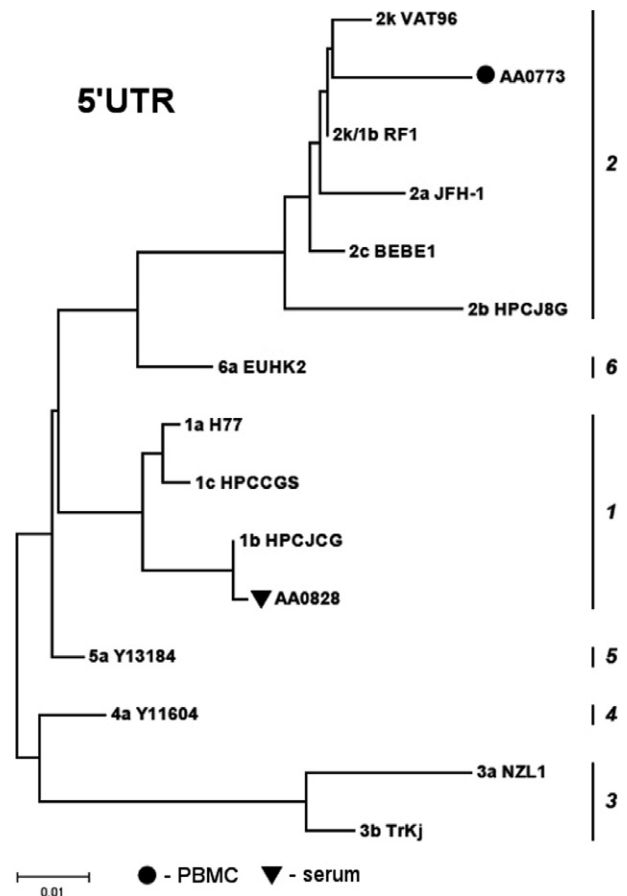
Conclusions: Development of side effects of the treatment with pegylated IFN-alfa-2a and ribavirin depends on some predictor factors.

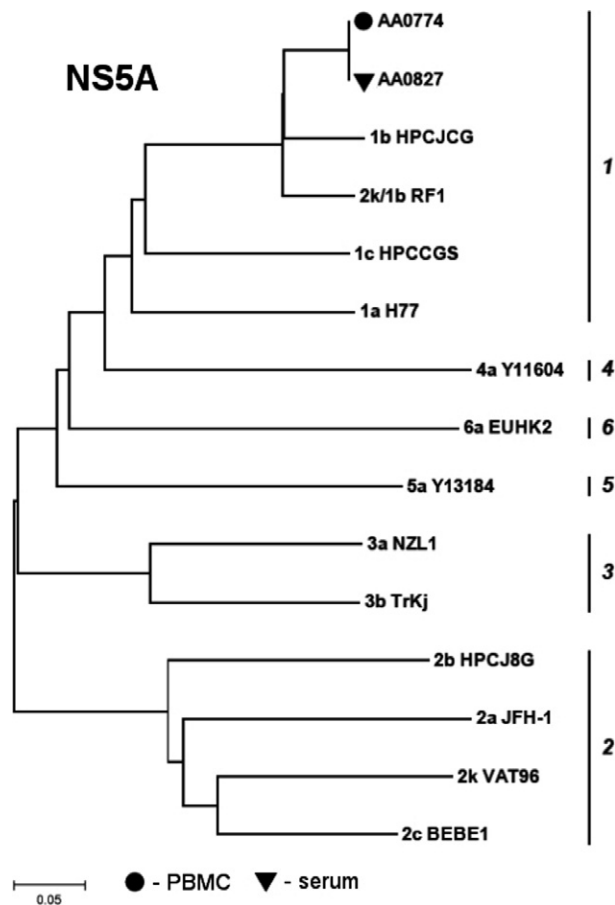
PP-145 Discordant results of HCV genotyping in peripheral blood mononuclear cells from patient with chronic hepatitis C: case report

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Introduction: There is a growing evidence of intergenotypic recombination in HCV. In this paper we report a patient infected by HCV 1b and, probably, recombinant 2/1b that is detected in peripheral blood mononuclear cells (PBMC).

Case description: Patient S., male, 31 years old admitted in January 2009. HCV viral load in serum before treatment – 9,630,000 IU/ml. HCV genotyping by sequencing 5'UTR and NS5A. According to phylogenetic analysis NS5A belongs to 1b (sera and PMBC), 5'UTR from serum – to 1b, from PBMC – to genotype 2. Due to discordant results recombinant 2/1b in PBMC can be suspected. NS5A interferon sensitivity determining region (ISDR) contains mutation R2218H. Laboratory: ALT 71 U/L, AST 62 U/L, GGT 36 U/L. Liver biopsy: HAI 8, fibrosis 1. Immunohistochemically HCV NS3 was detected in lobules and tracts. Elevated CD16 and CD20 was found in lymphoid follicles of portal tracts. Patient received treatment with peginteron (1.5 mg/kg BW) plus ribavirin (1000 mg/day) for 48 weeks. Virological and biochemical response were achieved on 12 wk and remained until the end of treatment and during follow-up. Liver biopsy after treatment: HAI 3, fibrosis 0. Immunohistochemically NS3 was still detected in lobules and tracts, CD16 and CD20 decreased in portal tracts.





PP-146 Establishment of highly efficient full-length HCV 1b genome cell culture system by inserting long distant structural fragment into replicon

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Objectives: To establish a full-length genome of hepatitis C virus (HCV) 1b, the dominant strain in China, cell culture system for further study.

Methods: The 5'-end of half HCV-1b genome (5.2kb) was amplified from Chinese chronic hepatitis C patients' seral samples with the refined long distant RT-PCR technique. The full length recombinant plasmid of HCV 1b was constructed by inserting the long distant structural regions directly into HCV 1b replicon containing the non-structural regions. In vitro transcribed genomic HCV RNA was transfected into Huh7.5.1 cells by liposome-mediated method. The real time quantitative RT-PCR, Western Blot, inoculation of naive Huh7.5.1 cells, immune fluorescence and titration of infectious HCV were used for identification of HCV replication and presence of infectious virion.

Results: The real time quantitative RT-PCR assay revealed the highest titer of HCV was 6.5×10^7 copies/mL in the cultural supernatants. While both Western Blot analysis and immune fluorescence confirmed the expression of HCV core protein in the transfected cells. Subsequent infection of naive Huh7.5.1 cells with supernatant of HCV cell culture resulted in high levels of HCV proteins and RNAs.

Conclusions: These results demonstrate the successful establishment of a HCV 1b culture system by the new strategy of inserting long distant structural amplicon into

replicon that produces infectious virus, which will allow the study of each aspect of the entire HCV life cycle and related studies.

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PP-147 Construction of a chimeric GB virus B with hepatitis C virus NS2-NS4A

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Background: HCV infection became a worldwide threat to human health, which can lead to chronic hepatitis C, cirrhosis and hepatoma. As only human and chimpanzees are susceptible to hepatitis C virus, the progress of HCV research has been obstructed due to the absence of a reliable small primate animal model. GB virus B (GBV-B), being very close to hepatitis C virus (HCV) phylogenetically, can replicate effectively in vivo of common marmosets, that has been made an attractive surrogate virus for HCV replication study. It was reported that HCV NS2-NS3 protease, NS3 protease and NS4A complex are critical for virus maturation and replication. The construction of chimeric GB Virus B with Hepatitis C NS2-NS4A region can be used to develop a marmoset model for antiviral and immune studies.

Methods: RT-PCR and overlapping PCR were applied to complete jointing gene fragments and T7 transcription kit was used to produce chimeric GBV-B/HCV infectious RNA in vitro.

Results: A chimeric clone originating from GB virus B (GBV-B), in which GBV-B NS2-NS4A region are replaced by analogous sequence of the HCV genome, was constructed.

Conclusions: This chimeric clone lays the roots for a surrogate model of Hepatitis C virus, insights into HCV replication mechanism and further HCV NS3 epitope-based research.

PP-148 Comparisons of molecular responses between recovery and chronic HCV infection from blood donors in Beijing and Guangdong, China

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Background: Hepatitis C virus (HCV) infection is a major public health problem in China. However systematic studies conducted on mechanism of recovery and chronic infection with HCV are little known. A detailed study was carried out to investigate IFN- α and IFN- γ correlating to antibody reactivity and viral factors in the population of recovered and chronic HCV infection from blood donors.

Methods: 160 plasmas samples reactive with anti-HCV assays were collected from blood donors in Guangdong and Beijing. HCV antibody reactivity was presented as S/CO by at least three EIA assays. All samples were tested for ALT and viral load, confirmed by nested-PCR, and classified as three statuses of recovery (RNA-/Ab+), chronic (RNA+/Ab+) and false positive (RNA-/Ab-) infections. Productions of IFN- α and IFN- γ in the serum were also quantified by ELISA. The genotypes of HCV from chronic samples were phylogenetically analyzed with 5'-NCR sequences (215-218bp).

Results: 45 recovery, 76 chronic and 39 false positive of 160 HCV antibody positive plasmas were finally confirmed. The rate of recovered HCV infected individuals in blood donors was 37.2% proximately. 63 HCV strains were genotyped,